

Contribution to the caryological study of the African grass *Aristida rhiniochloa* Hochst., based on specimens from the Southern Hemisphere

by

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ABSTRACT

The chromosome number established for *Aristida rhiniochloa* Hochst. by the study of material from three localities in the Southern Hemisphere confirms the results obtained on material from North Africa. The haploid complement ($n = 11$) and the diploid number ($2n = 22$) of this species conform to the basic number $x = 11$ typical for the Aristideae. It is shown that the karyotype of this species is sub-symmetrical. Preliminary studies of material from two localities show that the meiotic behaviour conforms to that found in the diploid species with n bivalents.

INTRODUCTION AND ACKNOWLEDGEMENTS

The study of *A. rhiniochloa* Hochst. in the Southern Hemisphere follows similar investigations started on North African material of this species, of which the distribution coincides for the greater part with the Sudan-Angolan phytogeographic region.

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METHODS

The study was partly done on material cultivated in a greenhouse in the Botanical Garden of the "Centre de St.-Jérôme" of the University of Provence. The root tips of plants (originally from Angola), were fixed in bichromate of potassium and stained in crystal violet (3). The young panicles of plants (originally from Rhodesia) were fixed in formula 3 of Carnoy (9) and stained in carmine-haematoxylin (1, 2, 5 and 9). The method of application of heat as suggested by Cauderon (10) was used. It is similar to the method usually applied by us, but the material is pre-stained at room temperatures and is only heated over a spirit lamp after squashing in 45% acetic acid or in acetic haematoxylin (2) diluted to 50%. Boiling the liquid must be avoided by passing the slide over the flame fairly rapidly, otherwise chromosome damage may result. Staining proved to be good with both methods and it was possible to study and photograph divisions after more than two months storage in a refrigerator. Slides stored in this way should be repeatedly passed above the flame of a spirit lamp immediately before study of the divisional plates under the microscope. This technique shows the colourless spindle clearly in prometaphase I and metaphase I of the divisions of the microspores (Fig. R1e). The photographs of the prometaphase and metaphase plates were taken by focussing on the chromosomes not the whole spindle. A beautiful spindle-vestige in telophase I of a pollen mother cell is visible in Fig. R1g.

RESULTS

Caryology of the Angolan material

Origin of material: Mucope (Lat. S., $16^{\circ} 25'$, Long W., $14^{\circ} 50'$), Angola. Collected by O. J. Azancot de Menezes. Date of collection: 1968.

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The chromosome number of $2n = 22$ was established and confirmed on five different metaphases found in root tip material. (See Fig. A.) The length of the chromosomes vary from $0,6-0,8 \mu$ for short and up to $1,4$ for long chromosomes.

Caryology of two collections from Rhodesia

Origin of material: Districts Wankie (near Victoria Falls) and Chiredzi. Collector: J. C. R. Hill. Dates of collection: August, 1969 and March, 1970 respectively.

1. *Chiredzi material*

In a study of pollen mother cells the number $n = 11$ was established 19 times with certainty on prometaphase I plates. (See Fig. R1c, R1d, R1f), four times on metaphase I plates (See Fig. R1e) and three times on anaphase I plates. The complement $2n = 22$ was established seven times on metaphases in ovaries. (See Fig. R1a and R1b). The length of the chromosomes varies from between $0,6$ to 1μ for the shortest chromosomes and $1,3$ to $2,8 \mu$ for the longest chromosomes.

2. *Wankie material*

Studies of pollen mother cells made it possible to establish the haploid number $n = 11$ with certainty on 22 diplotene plates, 91 times in diakinesis (See Fig. R2c), seven times in prometaphase I (See Fig. R2d, R2f, R2e) and six times in metaphase I. In the homeotypic mitosis of the meiotic divisions this haploid number was established 25 times with certainty on prophase II and five times on metaphase II. The chromosome complement of $2n = 22$ was established with certainty, at least three times on metaphases in the ovary (See Fig. R2b) and three times on metaphases in the stamens (See Fig. R2a). The length of the chromosomes vary from $1-1,3 \mu$ for the shortest to $2,1-2,8 \mu$ for the longest.

DISCUSSION OF THE CARYOLOGY

Both the haploid and diploid chromosome complements agree with the numbers established in somatic meristems of cultivated plants of this species from North Africa.

The caryotype

The study of the relative size and configuration of the chromosomes will be continued by at least one of us (P.B.) using more appropriate techniques, such as immersion, if necessary, in alphamonochloronaphthalene (10), treatment with pectinase i.e. rapidase C (17), staining in Feulgen followed by staining in acetic haematoxylin (9) or in acetic orcein (10). Methods of measuring used by Essad (18), Essad and Najcevska (19) will in addition be used. The measurements set out in Tables 1 and 2 can in the interim be supplied as an indication of the maximum and minimum lengths of the chromosomes treated with different fixatives.

TABLE 1.—Length of the longest and shortest somatic chromosomes of the root tips (Cr207K2 fixative).

Locality	(a) Length in μ of autosomes of minimum size	(b) Length in μ of autosomes of maximum size	"Ratio" $\frac{b}{a}$	Number of dif- ferent caryotypes measured
Attakou. Ennedi septentrional..	0,6	1,7	2,8	1
Airport El Obeid (Sudan).....	0,7	1,2	1,7	1
Airport El Fasher (Sudan).....	0,7-1	1,7-2	2,1	2
Angola.....	0,6-0,8	1,4	2,0	2

TABLE 2.—Lengths of the longest and shortest somatic chromosomes of the root tips (Carnoy type fixative).

Locality	Organ Studied	(a) Length in μ of the autosomes of minimum size	(b) Length in μ of the autosomes of maximum size	Ratio $\frac{b}{a}$	Number of different karyotypes measured
Airport of El Fasher (Sudan).....	basal meristem of leaf...	1, 2	2, 1-2, 6	1, 9	2
Chiredzi, District (Rhodesia).....	ovary....	0, 6-1	1, 3-2, 8	2, 5	7
Wankie, District (Rhodesia).....	stamen and ovaries...	1-1, 3	2, 1-2, 8	2, 1	2

An analysis of Tables 1 and 2 show that the relationship of the lengths of chromosomes of different pairs of autosomes varies from 1 to 2.8. The study of somatic metaphases, furthermore, showed that chromosomes may be distinguished as having meta- or submetacentric centromeres. The karyotype is therefore subsymmetrical. For a definition of the symmetrical karyotype the reader is referred to Stebbins (26).

If the width of the somatic chromosomes are compared it is seen that those of the roots are $\frac{1}{4}$ of the width of those of the leaves the stamens and the ovary. This may almost certainly attributed to the fact that two different fixatives were used.

The meiotic behaviour

The meiotic behaviour of the Rhodesian representatives is of the regular type found in diploid species (i.e. 11 bivalents in prophase I). The differences that exist are only those of relative frequency of the types of bivalents (II straight, II rings, II intermediates: angled, open rings). The frequency of the different types of bivalents and the chiasmata, will be supplied later in tables modelled on those used by Cauderon (10).

In prometaphase I and metaphase I certain bivalents are sometimes separated on the same spindle fibre (Fig. R1d and R1c). Such pseudo-univalents have in fact been observed in certain diplotene stages of two collections from Rhodesia. Our interpretation agrees with that given by Cauderon for a metaphase reduction division of *Agropyrum intermedium* (Host.) P. Beauv. (10) and by Geslot for certain metaphases I in the microspogenesis of *Campanula recta* Dul. (20).

In material from the Wankie district in nearly full prophase I a small spheroidal body, staining as deeply as the nucleolus and the chromosomes, was observed. It is not visible in any prometaphase, metaphase or anaphase plates, but it reappears in telophase. This cycle is shared by the nucleolus and it must therefore be considered as a nucleolus-satellite. This interpretation is supported by the presence of two nucleoli in some somatic cells. Although we have not examined meiotic prophase I in the material from the Chiredzi district, it seems probable that this peculiarity is characteristic of the species.

CONCLUSIONS

This study has confirmed that *Aristida rhiniochloa* Hochst. is a diploid with $2n = 22$ chromosomes and has contributed information on its karyotype and the meiotic behaviour of the diploids. The chromosome count $2n = 38$ for *Aristida rhiniochloa* in Darlington and Wylie (13) does not agree with our results and needs further investigation based on material of the species from the Sudan.

The present study is a preliminary one and should be extended to include a wider range of material to determine whether there are areas in which the species has developed polyploidy. Polyploid series in *Aristida* with 2-4 ploids (*A. fendleriana*, *A. glauca*, *A. longiseta*), 2-4-6-ploids (*A. wrightii*), 2-4-6-8 ploids (*A. purpurea*) have, as far as we know, for the first time been mentioned by de Lisle (14) in studies of material from the south-west United States.

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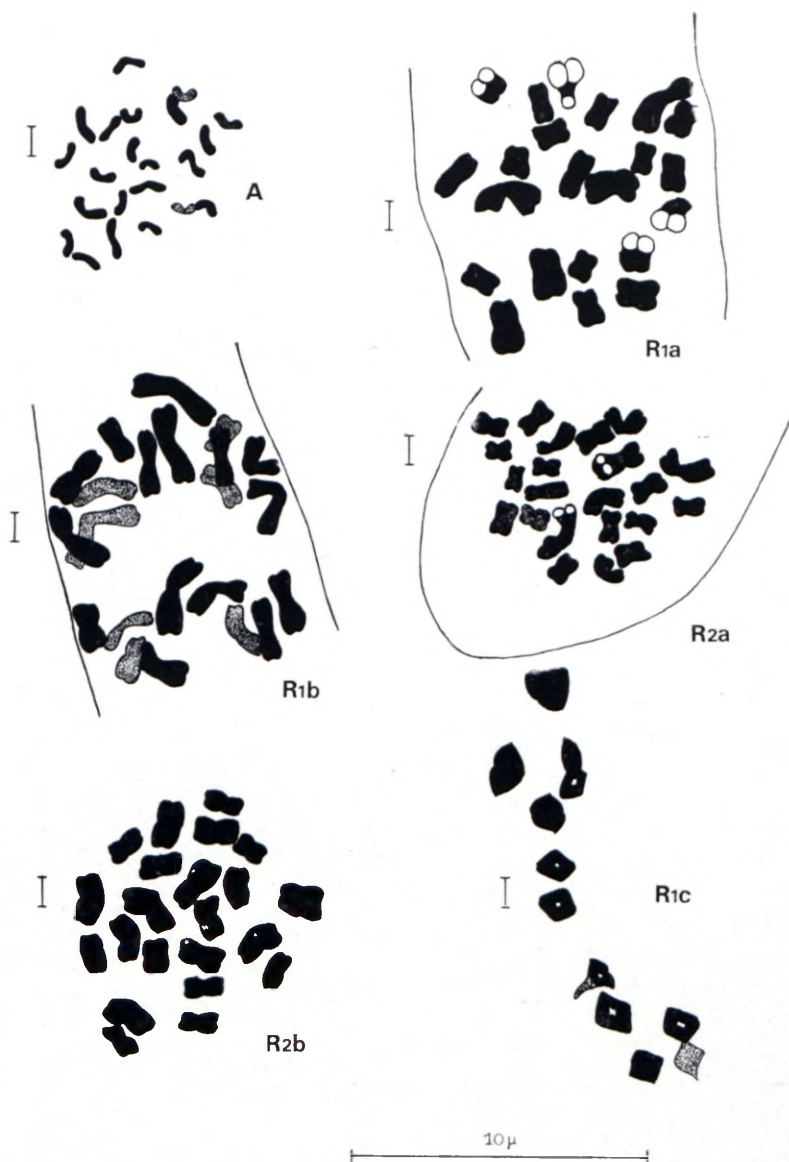


PLATE 1.—Drawings of the chromosome complement of *Aristida rhinochloa* obtained from cultivated plants.

Origin of the material: A, Angola. R, Rhodesia: R1, Chiredzi district; R2, Wankie district.

Explanation of the phases: Somatic metaphases ($2n = 22$) of: ovary (R1a, R1b, R2b); the stamens (R2a); the root meristem, (A). Meiosis: prometaphase I (R1c).

Type of association of the chromosomes.—Fig. R1c, 11. IIa (bivalents in rings, association very homogenous).

Note on the drawings. Certain chromosomes are drawn finely speckled to distinguish them from others with which they are in contact; vertical arms are outlined but left white. In heterotypic mitosis, drawing in black makes it difficult to give an accurate presentation of the chromosomes and each bivalent is drawn partially in black and partially in a hatching of black stipples. The vertical line to the left of each drawing represents 1μ .

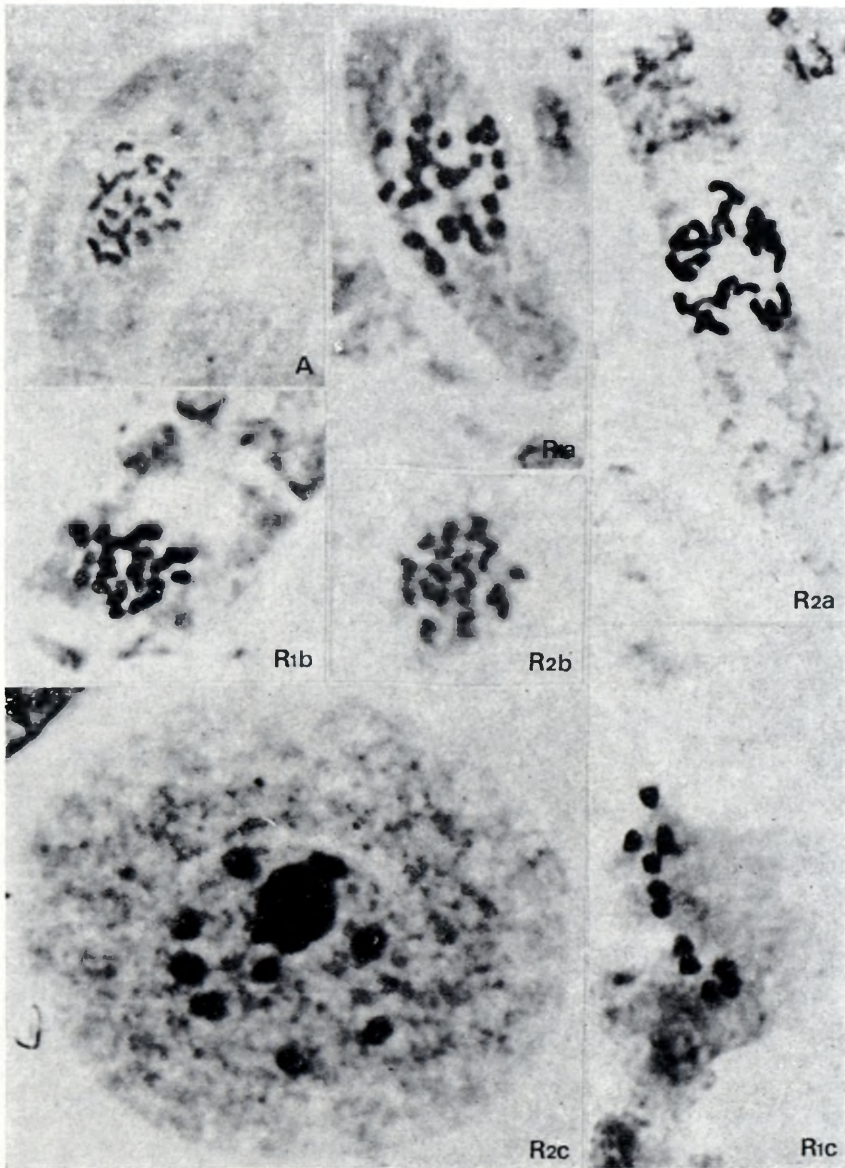


PLATE 1a.—Photographs of the chromosome complements of *Aristida rhinoclhoa*, which correspond with the drawings on Plate 1.

ERRATA—For R1b read R2a and for R2a read R1b.

N.B.—Note the absence in R1c of the membrane of the nucleus and nucleolus, as well as the absence of the achromatic spindle, also the bivalents which are aligned close together lengthwise in the cell corresponding to the position of the future spindle, not on the equatorial plane (characteristic of prometaphase I). Note in R2c (diakinesis, $n = 11$) the trace of the nuclear membrane and the deeply stained nucleolus which is much larger than the chromosomes.

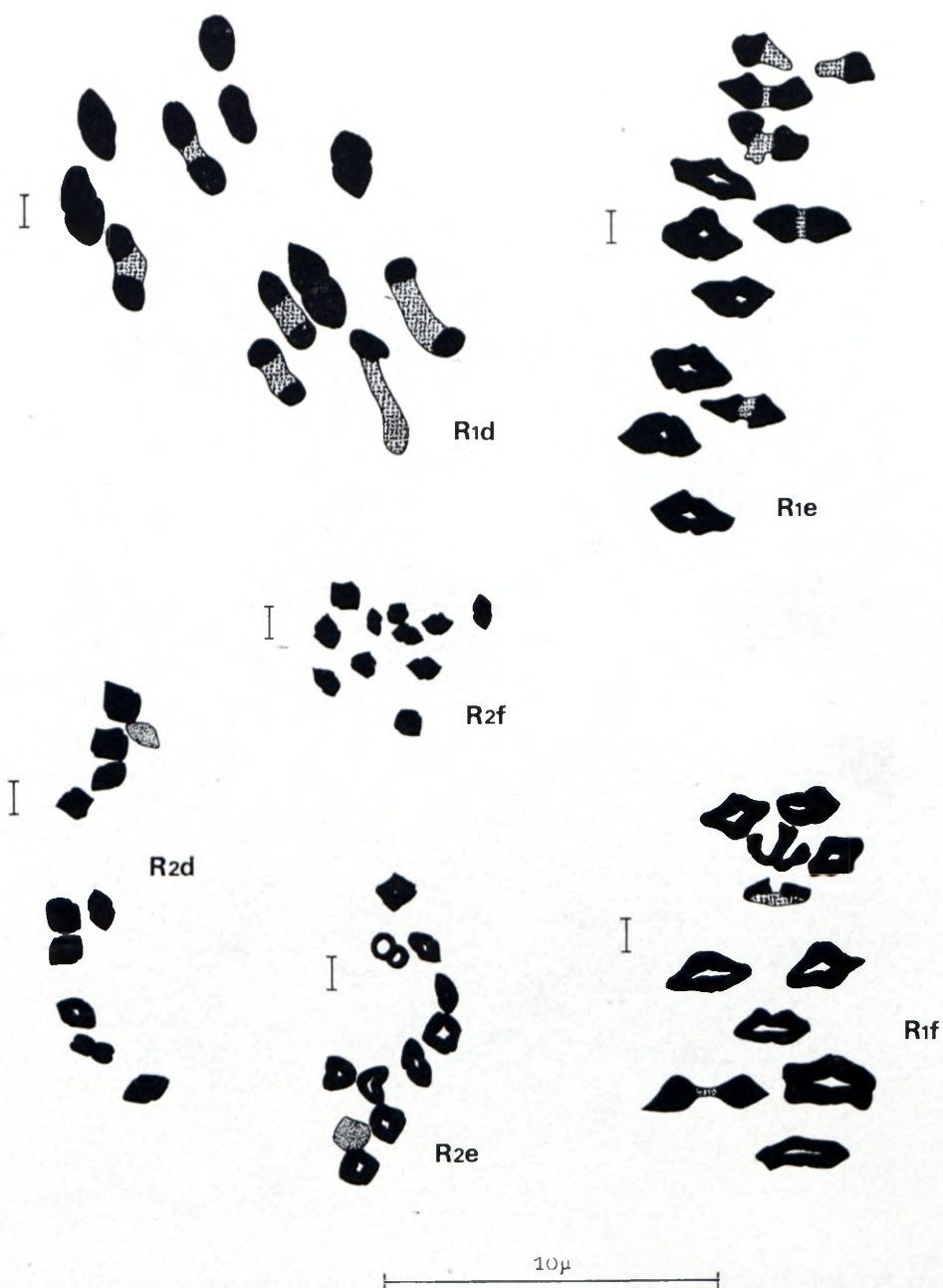


PLATE 2.—Drawings of the haploid chromosome complement ($n = 11$) of *Aristida rhiniochloa* obtained from cultivated specimens.

(For origin of material and notes on the drawing see Plate 1).

Explanation of the phases of the meiotic divisions of the pollen mother cells: Prometaphase (R1d, R1f, R2d, R2e, R2f); metaphase (R1e). N.B. In R1d the lowest bivalent forms a very open "elbow".

Type and association of the chromosomes:

- Fig. R1d, 4IIa + 1 IIa. dis. + 6 II dr.c. . . . = 11 bivalents (association less homogeneous).
 Fig. R1e, 6IIa. + 4 IIa.o. + 1 II dr.dis. . . . = 11 bivalents (association fairly homogeneous).
 Fig. R1f, 8IIa. + 3 IIa.o. = 11 bivalents (association homogeneous).
 Fig. R2d, 10 IIa. + 1II dr. = 11 bivalents (association fairly homogeneous).
 Fig. R2e, 11 IIa. = 11 bivalents (association very regular).
 Fig. R2f, 11 IIa. = 11 bivalents (association very regular).

Explanation of symbols: II = bivalent; a = a ring; a.o. = an open ring (the ring may be open from the start or more often the chiasmata may have been released in the twisting during prophase); a. dis. = ring separated into two chromosomes (pseudounivalents); dr. = straight; dr.c. = arms straight forming an "elbow"; dr. dis. = straight, separated.

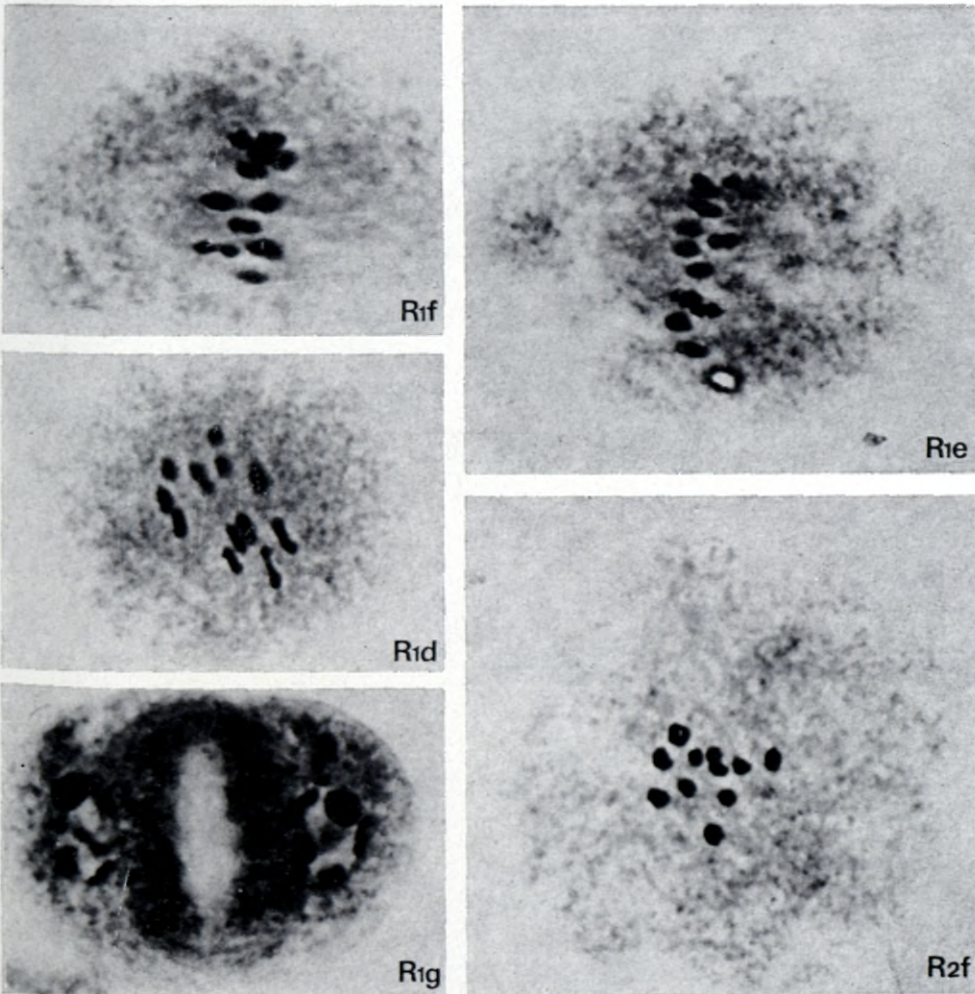


PLATE 2a.—Photographs of the haploid chromosome complements ($N = 11$) of *Aristida rhinochloa* corresponding to the drawings in Plate 2. Note in addition a telophase I (R1g).

